

# **RADIATION RETINOPATHY: AN EXPERIMENTAL MODEL FOR THE ISCHEMIC — PROLIFERATIVE RETINOPATHIES\***

*A. Rodman Irvine, MD, J. A. Alvarado, MD (BY INVITATION),  
W. M. Wara, MD (BY INVITATION), B. W. Morris  
(BY INVITATION), AND (BY INVITATION) I. S. Wood*

## **INTRODUCTION**

RADIATION RETINOPATHY IS BEING RECOGNIZED WITH INCREASING FREQUENCY BECAUSE OF THE INCREASING USE OF RADIATION IN OPHTHALMOLOGY. Many retinoblastomas and choroidal melanomas which formerly would have been treated by enucleation are now being irradiated.<sup>1-4</sup> In addition, there is a heightened awareness of the retinopathy following radiation to extraocular tumors.<sup>5-8</sup>

Radiation retinopathy is a vaso-occlusive process which induces ischemic and proliferative changes similar to those found in diabetic retinopathy.<sup>9</sup> The study of radiation retinopathy is, therefore, of interest not only in its own right but also as a possible experimental model for the other ischemic-proliferative retinopathies.

This paper reports the results of preliminary studies in the development of an experimental model for chronic vaso-occlusive radiation retinopathy in primates. These are long term experiments in which 2 to 3 years are required before the desired retinopathy is manifest. It therefore, seems worthwhile to report these initial findings so that others may consider using this model in their investigations of retinal vaso-proliferative factors.<sup>10</sup>

\*From the Department of Ophthalmology, University of California, San Francisco, CA. Supported in part by research grant No. EY 02162 from the National Institutes of Health and from the Department of Radiation Oncology, University of California, San Francisco, CA (Dr Wara).

## MATERIALS AND METHODS

## IRRADIATION AND CLINICAL OBSERVATION

Adult capuchin monkeys were anesthetized and their eyes irradiated with a 4 MeV linear accelerator. The monkeys were placed in restraining head holders and their position was checked with a television monitor throughout the treatment. The radiation dosage varied from 2,000 to 8,000 R, given in one treatment session from a single lateral port. A total of ten monkeys were radiated in four separate groups between November 1975 and August 1978 (Table 1). The monkeys were anesthetized with intramuscular ketamine and examined with indirect ophthalmoscopy every two to four months following radiation. Fundus photography and fluorescein angiography were performed whenever abnormal ophthalmoscopic findings were present as well as immediately prior to sacrifice.

## HISTOLOGIC STUDIES

Eyes in which radiation retinopathy was documented were studied histologically. In this pilot study, two of the animals which had developed retinopathy in the first treatment group (Table 1) died on a weekend following an examination under anesthesia. Their eyes were lost, and thus only five affected eyes were available for histologic examination. Three were in the early stages of retinopathy with only cotton wool spots, and two were in a later stage with what appeared angiographically to be early proliferative changes. In order to study these specimens by both conventional light microscopy methods as well as electron microscopy, they were divided in the following manner. Three eyes, two from the earlier and one from the later stages of the radiation retinopathy were used to perform conventional light microscopy studies. The other two eyes were prepared to be studied by electron microscopy, as described below.

The three eyes to be studied by conventional microscopy were fixed in formalin overnight. Then the eyes were sectioned with a razor blade along

TABLE I: TREATMENT GROUPS

TREATMENT SESSION	NO. MONKEYS	RADIATION DOSE	RESULTS
1	3	2,000-5,000 R	Chronic ischemic retinopathy
2	2	8,000 R	Acute retinal necrosis
3	3	3,500-4,000 R (Eyes not shielded)	Peforating corneal ulcers
4	2	2,500-3,000 R	Chronic ischemic retinopathy

the horizontal meridian leaving a central segment which included the pupil and the optic nerve as well as much of the macula. There was a superior and an inferior calotte from each eye sectioned. These two calottes were studied by means of digestion of the retina with trypsin as described by Cogan and Kuwabara. The central segment including the pupil and the optic nerve, was embedded in paraffin and sectioned in the usual, conventional manner and stained by hematoxylin and eosin.

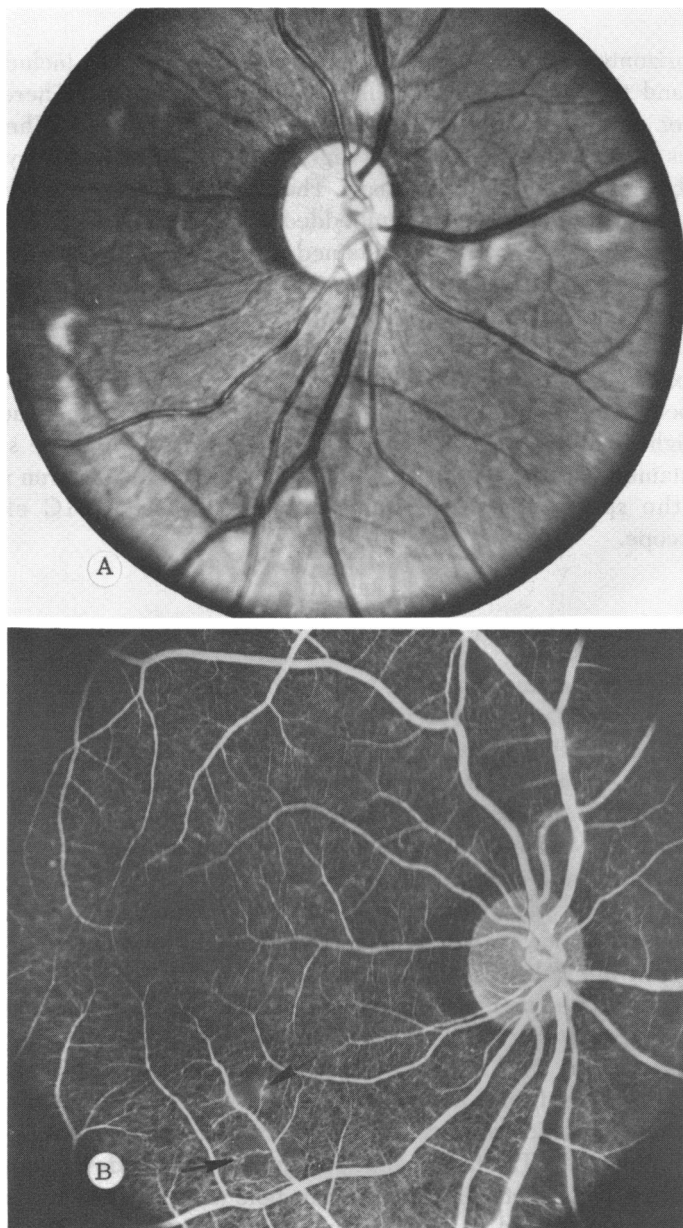
The two specimens used for electron microscopy were fixed for 24 hours in a combination of 2% paraformaldehyde and 1% gluteraldehyde buffered to a pH of 7.3 with Na cacodylate. The specimens were subsequently post-fixed in 1% of osmium tetroxide buffered in sodium veronal acetate. The specimens were embedded in Araldite and sectioned to be studied by both light and electron microscopy. For light microscopy, the sections were stained with a 1% solution of methylene blue. For electron microscopy the specimens were examined with a JEOL-101C electron microscope.

## RESULTS

### CLINICAL OBSERVATIONS

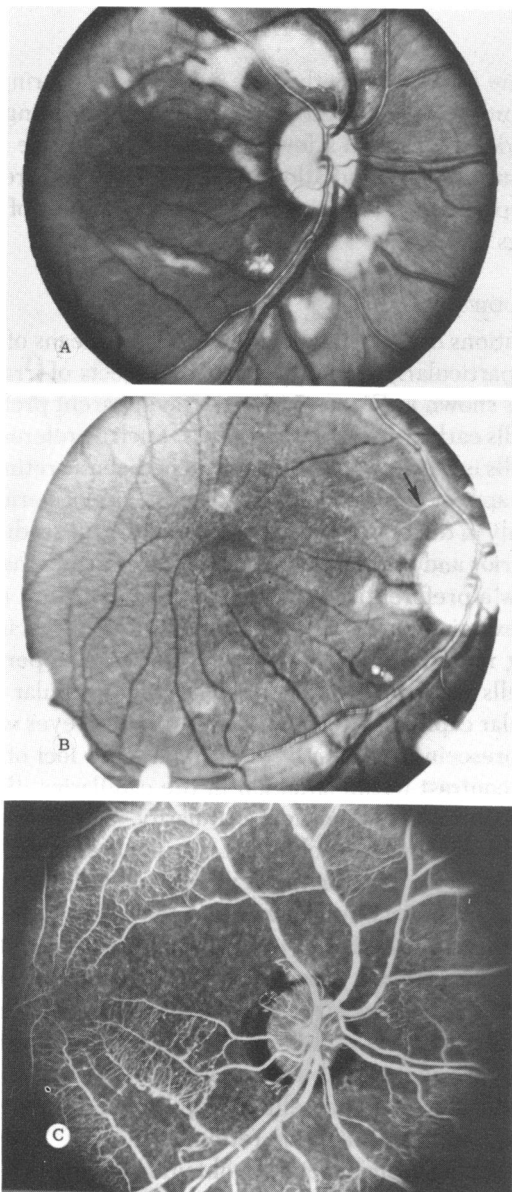
Of the four treatment groups, two failed to produce the desired vaso-occlusive retinopathy (Table I). One group failed because the anterior segments of the eyes were not protected with a lead shield. It was initially felt that the beam could be focused so as to miss the anterior segment, but in these small animals this proved false, and the radiation produced conjunctival drying and perforating corneal ulcers if the anterior segment was not protected with a lead shield directly adjacent to the eye. In another group, it was found that raising the radiation dose to 8,000 R did not produce a more rapid onset of the desired chronic vaso-occlusive retinopathy but rather caused an acute radiation retinopathy with massive photoreceptor necrosis, diffuse retinal vascular narrowing, and pigment epithelial atrophy.<sup>11-13</sup>

The remaining two treatment groups, totaling five treated animals, showed that doses from 2,500 to 4,000 R led to the development of a slowly progressive, chronic, vaso-occlusive radiation retinopathy identical to that seen in humans.<sup>5-9</sup> The first change recognized was the development of a few "cotton wool spots" in the posterior pole (Fig 1). The time of onset varied between 10 and 20 months from radiation, and within the 2,500 to 4,000 R range no correlation could be made between the dose and the time of onset of the retinopathy in these few animals. The number of cotton wool spots slowly increased, leading to large areas of retinal capillary non-perfusion (Fig 2). The cotton wool spots then regressed and the retinal



**FIGURE 1**

**Earliest radiation retinopathy. A:** Earliest sign of radiation retinopathy is the appearance of multiple cotton wool spots. **B:** Fluorescein angiography of same eye reveals small foci of capillary non-perfusion at the cotton wool spots with occasional persistent capillary "shunt" vessels similar to those in diabetes (arrow).



**FIGURE 2**

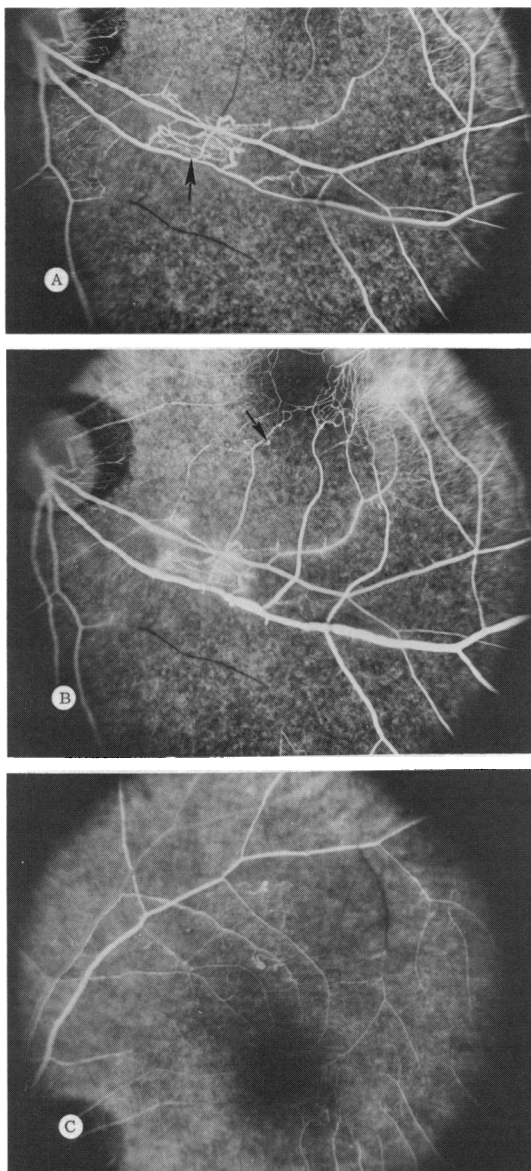
Later progression of radiation retinopathy to arteriolar closure with large areas of ischemic retina. A: 19 mos post irradiation, larger cotton wool spots and some hemorrhage have developed. B: 23 mos post irradiation, most of the cotton wool spots have resorbed, leaving white lines in place of the arterioles (arrows). C: Fluorescein angiography at 23 mos confirms that the white arterioles are occluded and outlines the large areas of retinal capillary non-perfusion.

arteries became attenuated with some arterioles appearing as thin white lines, while some veins showed dilation and mild beading. Some retinal hemorrhages developed and a few microaneurysms were seen. Approximately two years following radiation, small patches of apparent neovascular proliferans appeared at the margins of the large areas of capillary non-perfusion (Figs 3 & 4).

#### HISTOLOGIC STUDIES

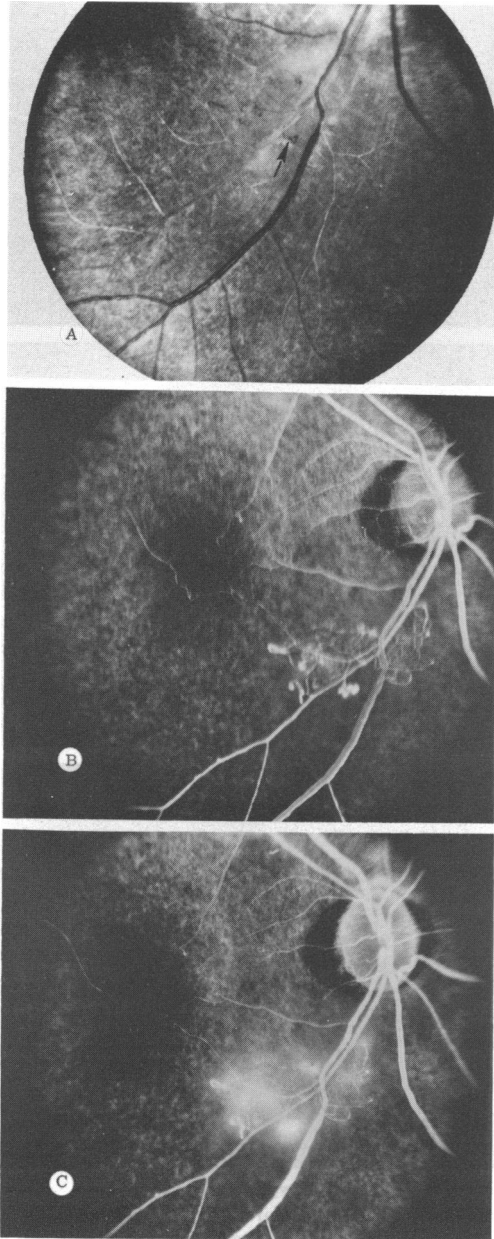
The preparations of the retinal blood vessels by means of digestion with trypsin were particularly useful to study the effects of irradiation on the lining cells. As shown in Figure 5, there is an apparent preferential loss of endothelial cells early in the disease process. Such a preferential damage to endothelial cells is contrary to that reported in diabetic retinopathy, where the pericytes appear to disappear first.<sup>14</sup> The loss of pericytes has been reported mainly in the posterior pole of diabetics. Our studies were mainly from the superior and inferior callotes, and it is possible that the posterior pole may show a preferential loss of pericytes.<sup>14</sup> Further, our studies are based on the examination of only a limited number of specimens. In other areas showing more severely affected capillaries, the pericytes and the endothelial cells were both absent, leaving thin, acellular strands. Large areas of acellular capillaries seemed to be present in eyes which had been shown by fluorescein angiography to contain large foci of capillary non-perfusion. In contrast to the situation in the capillaries, the venules and arterioles appear to have a normal cellularity. The preservation of the lining cells and coat in arterioles and venules persisted even in those vessels which were in the immediate vicinity of acellular capillaries. The trypsin digestion studies also showed that the capillary loss is multifocal and the distribution of such foci of abnormal vessels is rather irregular. Some of the areas affected showed only individual abnormal capillaries while other areas showed obstruction of a precapillary arteriole and all its associated capillaries. This multifocal pattern of capillary abnormalities is similar to that observed in diabetic retinopathy.<sup>15</sup> The striking scarcity of microaneurysms and of hypercellular capillaries, however, is in sharp contrast to the findings in diabetic retinopathy.<sup>16</sup>

Histologic sections from specimens obtained during the stage of cotton wool spot formation, early in the onset of the radiation retinopathy, show that the photoreceptors and the outer retina are relatively normal (Fig 6). Thereafter, the degenerative changes in the retinal non-vascular tissues seem to parallel those observed in the blood vessels. The blood vessel findings with electron microscopy are contrary to our interpretation of the findings from the trypsin digestion preparation. We found that the endothelial cells



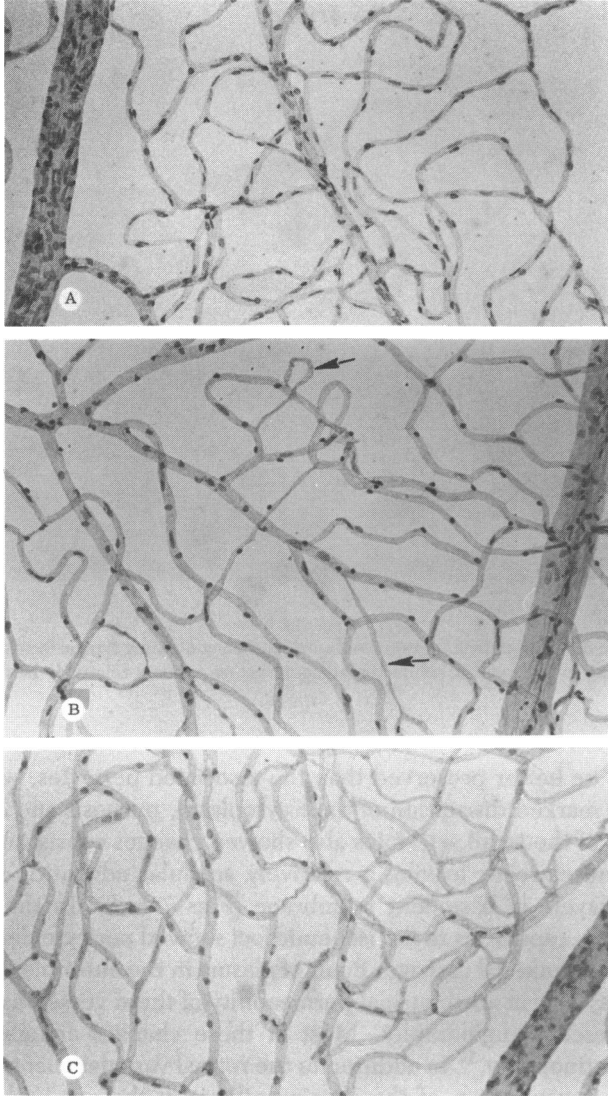
**FIGURE 3**

Intraretinal microvascular anomalies similar to those in diabetes. A: At 24 mos post irradiation, this eye has passed through the stages seen in Figs 1 and 2. Fluorescein angiography now reveals a group of markedly abnormal, dilated vessels within a large area of capillary closure (arrow). B: Later in the same angiography there is leakage from these abnormal vessels. A few microaneurysms can also be seen at juncture of perfused and non-perfused capillary net inferonasal to the fovea (arrow). C: Superior to the fovea more microaneurysms and a dilated tortuous capillary loop are seen.



**FIGURE 4**

**Apparent neovascular proliferans. A: 24 mos post irradiation this eye has marked arteriolar occlusion and shows apparent early buds of neovascular proliferans (arrow). B. and C: Fluorescein angiograms show the morphology and leakage of this apparent early proliferans.**



**FIGURE 5**

Preparation of retinal blood vessels by trypsin digestion. A: In control eye the capillaries have a ratio of endothelial cells to pericytes of approximately 1:1. No acellular capillaries were found (X100). B: Early stage of retinopathy, with a few cotton wool spots as the only ophthalmoscopic abnormality, scattered acellular capillary strands are seen in areas away from cotton wool spots (arrows). Note that endothelial cells seem to have lost their staining characteristics or dropped out (X100). C: Eye with more advanced retinopathy (seen in Fig 4), a small focal area where all the capillaries are acellular is seen in the upper right hand portion of the field, while adjacent capillaries show endothelial loss with relative sparing of pericytes. Elsewhere in preparation from this eye, much larger areas of totally acellular capillaries were present (X100).

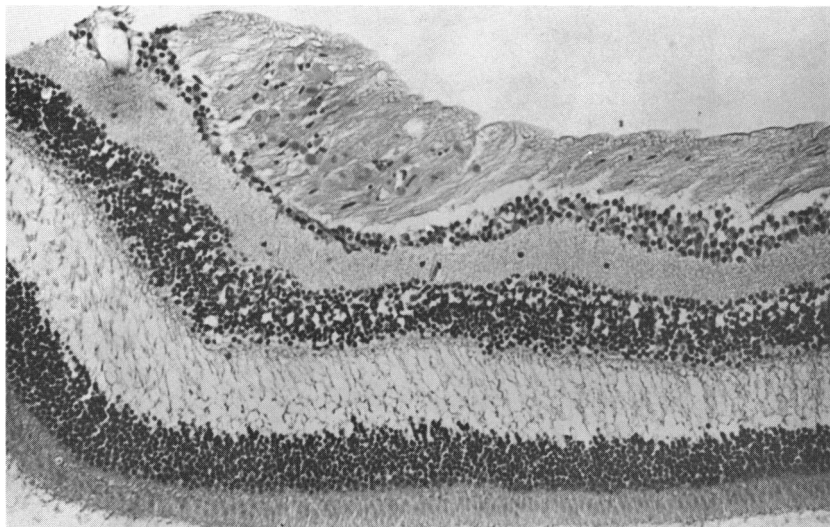


FIGURE 6

In early stage of retinopathy a "cotton wool spot" is seen containing typical focal area of axonal swelling and cytooid bodies, while the adjacent retina appears relatively healthy (X64).

appear to be better preserved than the associated pericytes, which often showed a marked disruption of their cytoplasm, pycnosis and cell death. The walls of the small arterioles also showed changes consisting in loss of smooth muscle cells leaving a relatively acellular adventitia containing multiple layers of basement membrane (Figs 7 & 8). In the advanced retinopathy, capillaries in the ischemic foci showed pericyte degeneration as well as endothelial damage. Pools of plasma in the outer plexiform layer (Fig 9) may reflect an abnormal permeability of these vessels as shown by our fluorescein angiography. Most of these changes are also seen in diabetic retinopathy.<sup>17</sup> In addition to the retinal vascular changes, we also observed degeneration of the choriocapillaris in the eye with the most advanced retinopathy (Fig 10).

In the two eyes which appeared to show early proliferative changes on fluorescein angiography, changes possibly consistent with intraretinal neovascular proliferation were found histologically, but none of these vessels were found extending through the internal limiting lamina onto the surface of the retina. The eye with the most convincing fluorescein angiographic evidence of proliferans (Fig 4) was fixed in formalin and studied by trypsin digest and routine thick sections. In the region of the lesion,

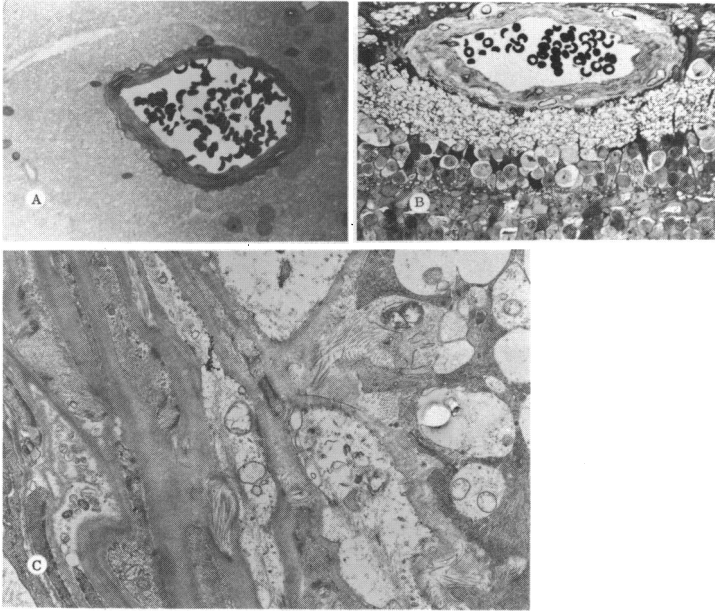
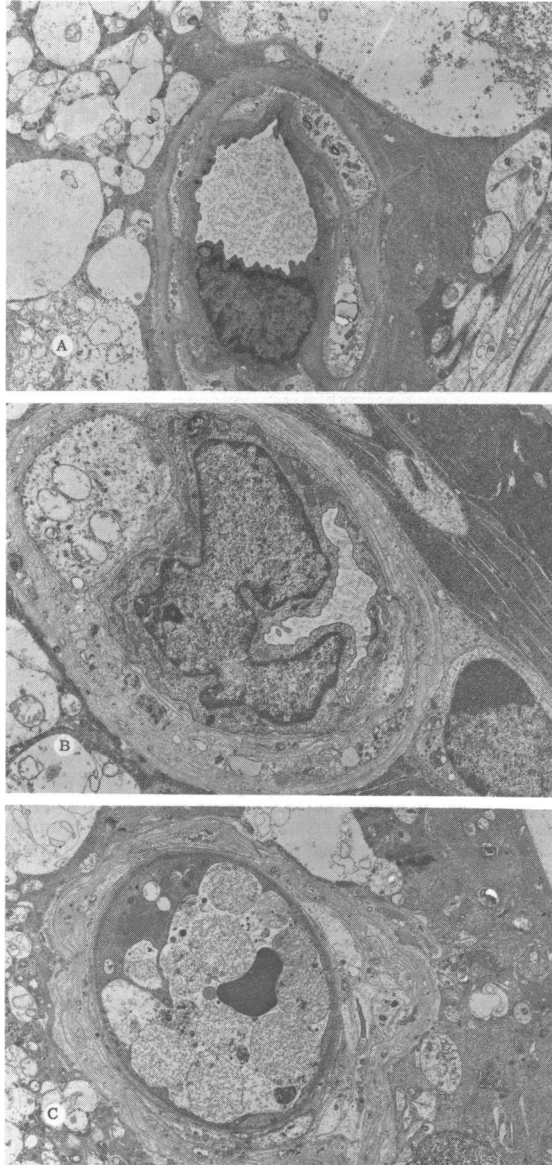


FIGURE 7

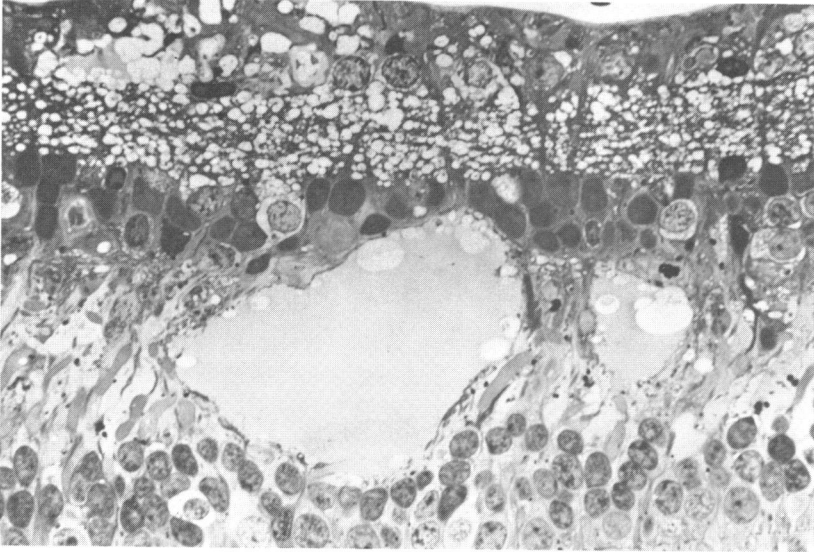
Arteriolar changes. A: Control eye, showing normal arteriolar wall with healthy muscular coat (X560). B: Arteriole shows loss of cells in vessel wall. Cells also show abnormal swelling and vacuolization of the cytoplasm of muscle cells (X560). C: Electron microscopy shows endothelial cell lining the lumen in upper right of the figure with a healthy appearance and normal organelles. Some smooth muscle layers, however, show a marked cytoplasmic degeneration while other smooth muscle cells are relatively intact (X12,960).

markedly abnormal, dilated vessels running just beneath the internal limiting lamina were found (Fig 10). The eye with microvascular changes appearing more similar to what has been termed "intraretinal microvascular anomalies (IRMA)" in diabetes (Fig 3), was studied with one micron sections and electron microscopy. The area of greatest microvascular anomaly revealed dilated, superficial vessels which had markedly abnormal walls. An extensive collagenous tissue surrounded the thin endothelial lining (Fig 11). The vessel lumen was quite large and enclosed by very thin endothelial cells. No fenestrae could be found in these endothelial cells. The abnormal collagenous tissue was reminiscent of that seen by Wallow and Gelder in a pre-retinal neovascular frond,<sup>18</sup> and by Archer and Gardiner in experimental subretinal neovascularization.<sup>19</sup>



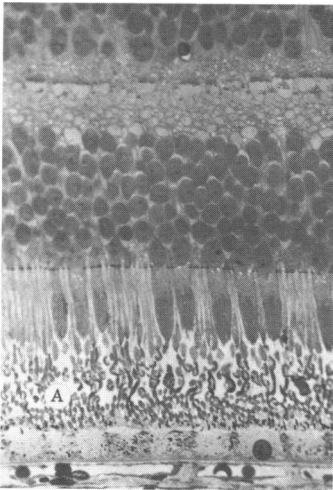
**FIGURE 8**

Capillary changes appear similar to those in diabetic retinopathy A: Endothelial cell looks relatively normal, whereas cytoplasm of pericyte shows striking vacuolization and loss of organelles indicating cell degeneration (X6,270). B: Capillary shows similar pericyte degeneration, while endothelial cell appears healthy. There is marked laminar arrangement of basement membrane (X9,500). C: This apparently occluded capillary was found in the region of some microaneurysms noted on fluorescein angiography. It has platelets filling its lumen and a markedly laminated basement membrane (X4,720).



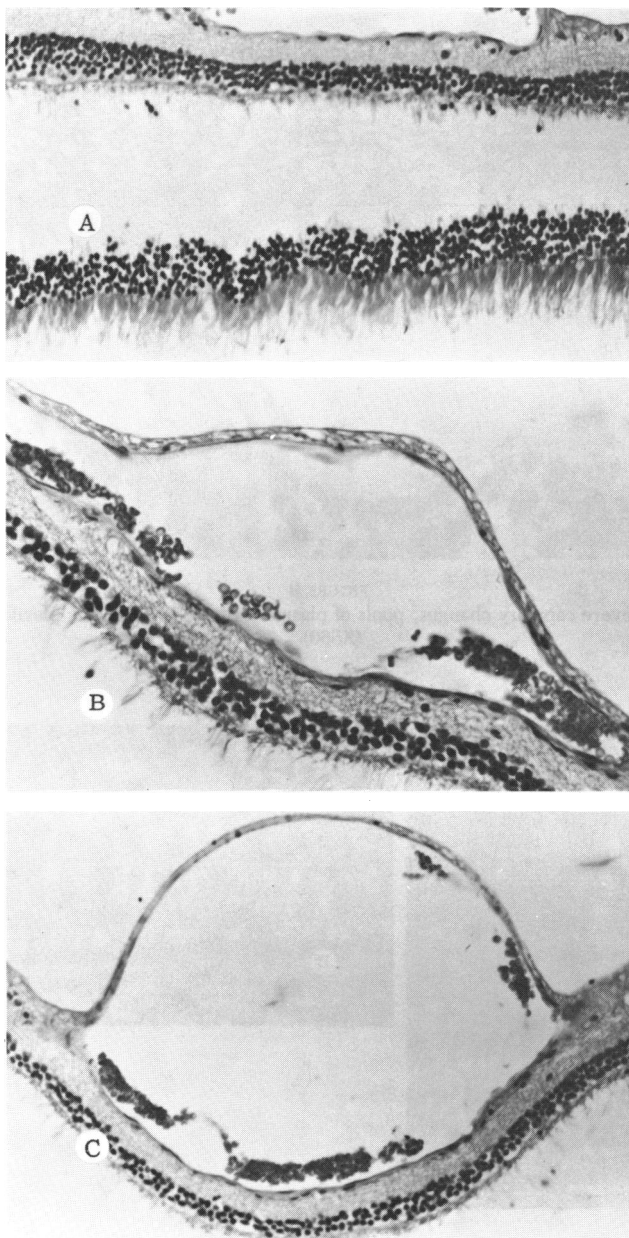
**FIGURE 9**

In areas of severe capillary changes, pools of plasma are seen in the outer plexiform layer. (X560).



**FIGURE 10**

Choriocapillary degeneration in a severely affected eye. A: Control eye shows a normal choriocapillaris and a healthy pigment epithelium (X560). B: Area of choriocapillaris atrophy where apparent "ghost vessels" have lost their cellular lining (arrow) (X560).



**FIGURE 11**

Abnormal vessels found histologically in area that appeared like neovascular proliferans on fluorescein angiography (Fig 4). A, B, and C: Following an extremely dilated, thin walled vessel. Although it develops marked aneurysmal dilatation (C) it remains below the internal limiting lamina (X140).

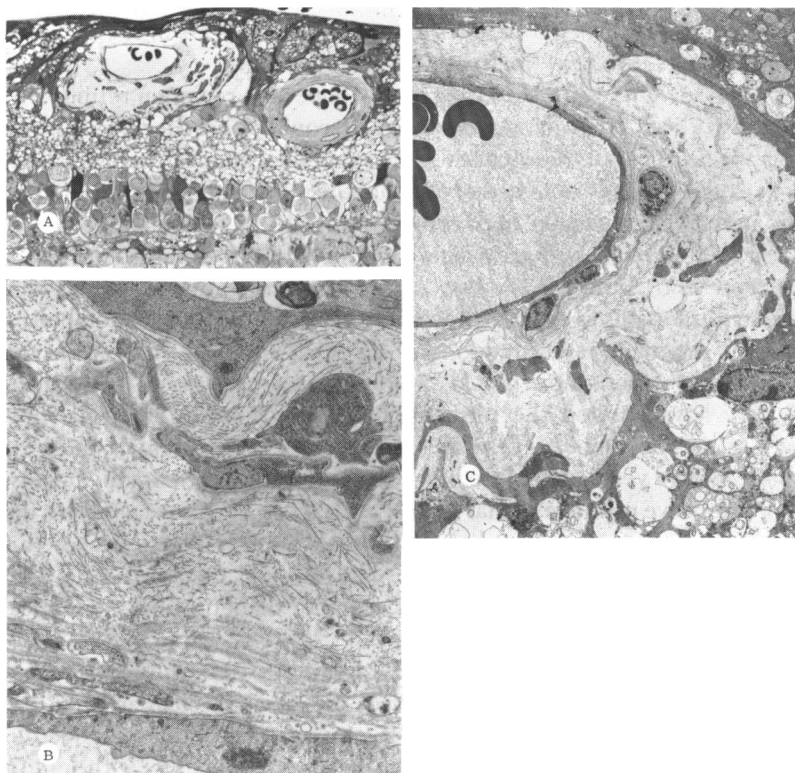


FIGURE 12

Abnormal vessels found histologically in area that appeared similar to intraretinal neovascular proliferans on fluorescein angiography (Fig 3A). A: Light microscopy reveals retinal arteriole with some degenerative changes in the cells of its muscular wall (vacuolation and loss of cytoplasmic staining) adjacent to an extremely abnormal vessel with a thin endothelial lining surrounded by a wide cuff of collagenous tissue (X560). B: Electron microscopy of the abnormal vessel in (A) shows the cuff of clear, collagenous tissue (X2,000). C: Higher power electron micrograph reveals apparently normal endothelial lining surrounded by collagenous tissue which is not present in adventitia of normal retinal venules (X15,770).

#### DISCUSSION

A reproducible experimental model for radiation retinopathy would be valuable for a number of reasons. It would make possible a controlled study of the effect of time-dose fractionation on the incidence of radiation retinopathy. Similarly the effect of the concurrent use of radiosensitizing drugs could be evaluated. Such studies are needed in order to determine

whether it is possible to separate the tumor killing effects of the radiation from its effects on the retinal vascular endothelium.

Of greater significance, however, is the hope that radiation retinopathy might provide an experimental model which could be used for a wide range of histologic studies, therapeutic trials, and biochemical investigations which would be applicable to diabetes and the other ischemic-proliferative retinopathies. Engerman and co-workers have developed a model of diabetic retinopathy in dogs which has proven of great value, but it has important limitations. With this model they have demonstrated that diabetic retinopathy is a product of the abnormality in glucose metabolism and not a separate, associated genetic defect. They have shown that the retinopathy develops more quickly when the diabetes is poorly controlled, and they have studied the ultrastructural basis for the abnormal permeability of the diabetic vessels. Unfortunately, the animals must be diabetic for five years before the first non-proliferative retinopathy begins, and proliferative retinopathy has not yet been seen. The time factor and the difficulty in maintaining the diabetic animals for this period makes this model impractical for most laboratories. In addition, the animals develop severe cataracts within two years of becoming diabetic, making clinical evaluation of the retinopathy impossible.<sup>20,21</sup>

In the radiation model, the stages of progressive multifocal retinal vascular closure and the development of proliferative changes could be followed ophthalmoscopically and then studied with light and electron microscopy on a large number of optimally fixed specimens with fundus photographic and fluorescein angiographic correlations. One would hope from such studies to develop a clearer understanding of pathogenesis. The vitreous could be assayed for vaso-proliferative factors produced by the ischemic retina. This could be done before and after various regimens of photocoagulation or cryocoagulation. Also, one could determine whether lensectomy and vitrectomy promoted rubeosis irides in these animals. If it did, the development of rubeosis irides might be used as an index to test the effects of chemical inhibitors of the vasoproliferative factor.

The present model appears as though it may be suitable for the above purposes. After 12 to 24 months, a vaso-occlusive retinopathy develops. It gradually progresses, producing large areas of ischemic retina. In the few eyes studied histologically, the changes were remarkably similar to those described in diabetic retinopathy. Only two eyes developed what appeared to be early neovascularization on fluorescein angiography, and histologic studies to date have shown evidence of only intraretinal proliferation with no proliferation through the internal limiting lamina. It seems likely that more proliferative retinopathy would have developed if the monkeys had

been followed longer. On the other hand, the relative scarcity of microaneurysm formation and the apparent preferential loss of endothelial cells in the trypsin digest preparations (limited to the areas peripheral to the vascular arcades in our study) might indicate that the radiation has damaged the endothelial cells throughout the retina so that they are incapable of the marked proliferative response seen in diabetes. In this respect, the model could possibly be more similar to central retinal vein occlusion, where the severe diffuse endothelial damage is felt to prevent retinal vascular proliferation.<sup>22</sup> More studies are needed to determine this. Even if it should prove that the retinal proliferans is restricted in this model, the ischemic stimulus is present and presumably the vasoproliferative chemicals. Also, since the anterior segment is shielded from radiation, rubeosis irides might prove a useful measure of the vasoproliferative stimulus.

It seems quite likely that this experimental model will prove applicable to a variety of studies. We, therefore, present these preliminary results in hopes that others will find the model useful.

#### ACKNOWLEDGEMENTS

Dr Robert Neger, while in his ophthalmology residency, helped greatly in getting the initial stages of this study underway.

Winifred Slauson prepared the trypsin digest studies and the serial routine light microscopy sections.

#### REFERENCES

1. Egbert PR, Donaldson SS, Moazed K, et al: Visual results and ocular complications following radiation for retinoblastoma. *Arch Ophthalmol* 96:1826, 1978.
2. Bedford MA, Bedotto C, MacFaul PA: Radiation retinopathy after the application of a cobalt plaque: Report of three cases. *Br J Ophthalmol* 54:505, 1970.
3. Char DH, Lonn LI, Margolis LW: Complications of cobalt plaque therapy of choroidal melanomas. *Am J Ophthalmol* 84:536, 1977.
4. Gragoudas ES, Goitein M, Verhey L, Munzenreider J, Suit HD, Koehler A: Proton beam irradiation: An alternative to enucleation for intraocular melanomas. *Ophthalmology* 87:571, 1980.
5. Chee PHY: Radiation retinopathy. *Am J Ophthalmol* 66:860, 1968.
6. Hayreh SS: Post-radiation retinopathy: A fluorescein fundus angiographic study. *Br J Ophthalmol* 54:705, 1970.
7. Bagan SM, Hollenhurst RW: Radiation retinopathy after irradiation of intracranial lesions. *Am J Ophthalmol* 88:694, 1979.
8. Tamsak RL, Smith JL: Radiation retinopathy in a patient with lung carcinoma metastatic to the brain. *Ann Ophthalmol* 12:619, 1980.
9. Gass JDM: *Stereoscopic Atlas of Macular Disease: Diagnosis and Treatment* (2nd ed). St Louis, CV Mosby, 1977, pp. 276-277.
10. Patz A: Studies on retinal neovascularization. *Invest Ophthalmol* 19:1133, 1980.

11. Cibis PA, Noell WK, Eichel B: Ocular effects produced by high intensity x-radiation. *Arch Ophthalmol* 53:651, 1955.
12. Cibis PA, Braun DV: Retinal changes following ionizing radiation. *Am J Ophthalmol* 49 (5II): 84, 1955.
13. Newton JC, Barsh-Newton MC, Warley J: The effects of x-radiation on the retina of the albino rabbit viewed with the scanning electron microscope. *Rad Research* 81:311, 1980.
14. Cogan DG, Kuwabara T: The mural cell in perspective. *Arch Ophthalmol* 78:133, 1967.
15. Bresnick GH, Engerman R, Davis MD, De Venicia G, Myers FL: Patterns of ischemia in diabetic retinopathy. *Trans Am Acad Ophthalmol Otolaryngol* 81:694, 1976.
16. Kohner E, Henkind P: Correlation of fluorescein angiogram and retinal digest in diabetic retinopathy. *Am J Ophthalmol* 69:403, 1970.
17. Ashton N.: Vascular basement membrane changes in diabetic retinopathy. *Br J Ophthalmol* 58:344, 1974.
18. Wallow, IHL, Geldner PS: Endothelial fenestrae in proliferative diabetic retinopathy. *Invest Ophthalmol Vis Sci* 19:1176, 1981.
19. Archer DB, Gardiner TA: Morphologic, fluorescein angiographic and light microscopic features of experimental choroidal neovascularization. *Am J Ophthalmol* 91:297, 1981.
20. Engerman RL: Animal models of diabetic retinopathy. *Trans Am Acad Ophthalmol Otolaryngol* 81:Op-710, 1976.
21. Wallow IHL, Engerman RL: Permeability and patency of retinal blood vessels in experimental diabetes. *Invest Ophthalmol* 16:477, 1977.
22. Chan C, Little HL: Infrequency of retinal neovascularization following central retinal vein occlusion. *Ophthalmology* 86:256, 1979.

## DISCUSSION

DR ARNALL PATZ. Doctor Irvine and co-workers are to be congratulated on the development of this experimental model of radiation retinopathy. It is an important contribution in that they have used a primate model, and furthermore the technique is reasonably reproducible and does not involve an invasive procedure.

The authors have carefully documented their findings by serial retinal photography and fluorescein angiography. The eyes were subsequently examined by both light and electron-microscopy and trypsin digest preparations of the retinal vessels. The radiation dosage varied from 2,000 to 8,000 R in these studies. It was of interest that with the high dose, instead of producing a more rapid onset of vaso-occlusive retinal vascular changes, the authors found massive photo-receptor necrosis, diffuse retinal vascular narrowing, and atrophic pigment epithelial changes without classic radiation retinopathy. The five treated animals receiving doses from 2,500 to 4,000 R showed classical radiation retinopathy as observed in humans. These included the original appearance of soft exudates (cotton-wool spots) slowly increasing areas of retinal capillary closure (non-perfusion with fluorescein), attenuated arteries, and dilatation with mild beading of some of the veins. Occasional retinal hemorrhages and microaneurysms were noted. Small patches of "apparent neovascular proliferans" appeared at the margins of large areas of capillary non-perfusion.

The trypsin digestion preparations were of interest as they showed varying degrees of endothelial cell damage and pericyte loss. In the monkeys with advanced retinopathy, capillaries in the ischemic zones showed both pericyte degeneration and endothelial damage. In the eyes with the most advanced retinal changes, the choriocapillaris showed degeneration.

The neovascularization observed was confined histologically within the substance of the retina and the authors have properly identified these as "intraretinal neovascular proliferation."

The authors' studies represent an important contribution in two separate areas. First, precise quantitation of the retinal vascular side effects of "tumor killing" radiation can be examined in this model. Second, and possibly of greater importance, is that the studies provide an experimental model applicable to all of the ischemic-proliferative retinopathies, particularly diabetic retinopathy. The model would be quite useful for the study of vaso-proliferative or angiogenesis factors once the retinal vascular occlusion has developed. The authors also indicate the possibility of the testing of vaso-proliferative inhibitors in this model. Most importantly, the authors have provided a primate model for experimental study. It is significant that their present studies show only "intra-retinal retinopathy" in contrast to the intravitreal retinopathy produced in the oxygen model of retrolental fibroplasia in young animals. The authors have postulated that with a longer period of follow-up some of the intra-retinal neovascularization could have proliferated through the internal limiting membrane. This is a logical suggestion and it is quite possible that with longer follow-up intra-vitreous neovascularization will develop.

Again, the authors are to be congratulated on this excellent study. Their experimental model has the potential of advancing significantly our understanding of the basic mechanisms in the development and control of retinal neovascularization.

DR WALLACE McMEEL. This is a very exciting topic. Using radiation in combination with the production of the chemical diabetes (ie, alloxan or streptozotocin) might be a very good way of producing proliferative retinopathy.

DR THOMAS R. HEDGES. I too would like to congratulate the authors for a remarkably well worked out study. Those of us who are in neuro-ophthalmology are very familiar with the devastating results of radiation treatment to the optic nerve and chiasm, especially in acromegaly or in patients with invasive pituitary adenomas that extend beyond their normal confines and cannot be removed by transphenoidal surgery. I would like to ask the authors the following: (1) Have they studied the optic nerves in these animals? and (2) whether they have in their review of the literature any further information with regard to the integrity of the optic nerve in a study of this type. Again my congratulations.

DR ROBERT W. HOLLENHORST. I have really enjoyed this paper as I have been interested in radiation retinopathy for a long time. Back 25 years or so, when we were still irradiating pituitary tumors, occasionally we would see a patient or two with microaneurysms of the retina but with no explanation for them. At that time we had no idea why these developed. I hope the authors will carry on their investigations to establish the minimal dose that will produce reactions of this type in the retina. Clinically, in our patients it was difficult to estimate how much radiation the eyes received. No matter how much the eyes are shielded and how remote the target site, there are distinct potentialities for damage, as Doctor Bagan

and I demonstrated in our paper two years ago. These fundus photographs are from a patient who had an inoperable frontal lobe tumor treated with 7200 rads in 1975. (slide) No retinal damage was evident in 1977, two years later, but in 1978 he had as you can see numerous tiny little microaneurysms in both eyes. (slide) These had decreased in number by 1979 and had lessened even more by 1980, however mild posterior subcapsular lens opacities were first observed five years after treatment was given.

The next patient had 7200 rads of Cobalt 60 in 1970 for a squamous cell epithelioma of the right maxillary sinus. She was first examined ophthalmoscopically five years later and at that time she manifested small hemorrhages, cotton wool patches, and microaneurysms only in the ipsilateral eye. Four years later she had a massive vitreous hemorrhage from a vascular loop. She had this only in the one eye. The other eye was free of problems.

The last patient, whose retinas are illustrated, had the worst radiation retinopathy that I had ever seen, in both eyes. (slide) Ten years before I saw him he had had treatment with  $^{31}\text{I}$  for thyrotoxicosis. Four years before I saw him he developed a malignant exophthalmos treated with an unknown dosage of Cobalt 60 in the Phillipine Islands. The retinopathy was very similar in the two eyes but I show only the one eye, illustrating the severe involvement of the optic disc and of the retina. Six months later the process had resolved somewhat. However, he ended up with only light perception vision in both eyes.

DR A. RODMAN IRVINE. I thank you all. I am thrilled, Dr Patz, to think that you might turn the techniques and bioassays that your group has developed onto our model. I hope it will prove of value.

Dr McMeel's idea is helpful. All of us have seen how hypertension and diabetes seem to synergize and make the process go faster. In our radiation model, we've waited two years for the retinopathy to develop, and we certainly would like a way to make it move faster. Frankly, I had not thought of combining it with diabetes. This sounds like a good idea. We tried to increase the radiation dose to see if that would make the process move faster. In fact, however, if you increase that dose then you get into a radiation retinopathy of an entirely different type. You get severe, immediate radiation damage to the photoreceptors, and you get a picture that looks like retinitis pigmentosa developing within three months of radiation treatment. It's an entirely different picture; so in order to develop the chronic ischemic retinopathy you have to be down in the range of treatment below where you destroy the photoreceptors.

Doctor Hedges asked about the optic nerve, and I really have to say at this stage we haven't done good studies on the nerve. We've looked at the nerve immediately behind the globe and seen a marked loss of axons, but we haven't studied the vessels in the optic nerve. Most of the work that others have done on radiation damage to the brain and optic nerve has dealt with acute changes due to high dose levels, little has been done on the chronic vascular effects of lower doses. We still have a lot to learn, and that's why we will have to rely on Doctor Alvarado to interpret the histologic and electron microscopic findings.